

Serological profile of some viral infections in unvaccinated cattle in Turkey

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Summary

In this study, 936 unvaccinated cattle sera were analyzed against five viruses: bovine viral diarrhoea virus (BVDV), parainfluenzavirus type 3 (PIV-3), bovine adenovirus type 1 (BAdV-1), bovine adenovirus type 2 (BAdV-2) and bovine adenovirus type 3 (BAdV-3), which were caused by respiratory system diseases in cattle. The study used the serum neutralization (SN) test – a conventional method. The seropositive rates for each virus were found to be 20.19%, 41.02%, 23.82%, 21.90% and 8.1%, respectively. Evaluation of the data revealed that 25.64% of the sampled population was seronegative against the investigated viruses, 74.36% were seropositive for one or more of them and 42% of animals were found to be seropositive against any of the investigated viruses. 25.42% of the animals were seropositive against two viral infections whereas 5.23% of the animals were found to be seropositive against three viral infections and 0.1% of the animals were detected to be seropositive for four viruses.

Keywords: Bovine viruses, respiratory system

Small scale cattle breeding has economical importance for many people living in rural areas in Turkey. Currently, viral infections causing respiratory diseases are the major health problems in cattle breeding. They are also responsible for considerable economic losses. Bovine viral diarrhoea virus (BVDV), parainfluenzavirus type 3 (PIV-3), and bovine adenovirus types 1, 2 and 3 (BAdV-1, 2, 3) are important agents causing respiratory tract infection and are frequently detected in cattle (1, 2, 12). These viral agents may cause acute or subacute infections which reduce meat and milk production in cattle.

BVDV, a member of the *Pestivirus* genus of the *Flaviviridae* family, is widespread throughout the world. Infection with BVDV has a wide clinical spectrum that varies from a mild and moderate subclinical form to the highly fatal form known as mucosal disease (3, 7, 8). Transplacental infection may occur during pregnancy and fetuses infected early may develop immunotolerance and persistent infection (PI). These animals are the major source of the infection (11, 14) PIV-3 is a member of *Paramyxovirus* genus of the *Paramyxovirinae* subfamily of the *Paramyxoviridae* family (3). This agent causes subclinical infection and clinical symptoms arise in the presence of secondary pathogens (13). BAdVs are also important respiratory pathogens and cause acute or subacute viral disease in

cattle that is characterized by pyrexia, nasooocular discharge and pneumonia (1). These agents are members of the genus *Mastadenovirus* of the family *Adenoviridae* (9).

To elicit further information, the current study was undertaken and we hereby report the results of the serological profile of some viruses causing respiratory disease in unvaccinated cattle from six herds in three provinces of Turkey.

Material and methods

Serum samples. Blood samples were collected from a total of 936 cattle from six herds located in three different provinces of Turkey (Samsun, Tokat, Sivas) and transported to the laboratory at 4°C. The samples were centrifuged at 3000 rpm for 15 min. at 4°C. The sera were subsequently separated into Eppendorf vials, heat inactivated at 56°C for 30 min. and stored at –20°C until tested. Sampled animals were 2 to 3 years old and unvaccinated against the investigated viruses.

Viruses and Cell culture. BVDV (NADL strain), PIV-3 (SF4, German strain), BAdV-1 (strain 11/66), BAdV-2 (Strain 12/66) and BAdV-3 (strain 13/66) were used in this study. BVDV free MDBK (Madin Darby Bovine Kidney) cells maintained with Dulbecco's minimal essential medium (DMEM, PAA, Inc, Austria) containing between 2% to 10% foetal calf sera (FCS, PAA Inc, Austria) were used for propagation, titration and virus neutralization tests.

Microneutralisation tests.

The virus neutralization (VN) protocol described by Frey and Liess (6) was performed using MDBK cells in 96 well microplates (Grainer, Germany). Fifty μ l of each serum was added to wells, mixed with an equal volume of 100 TCID₅₀ dilution of the selected viruses and incubated for 1 h at 37°C. MDBK cells were then distributed into wells. Test plates were incubated at 37°C in 5% CO₂ atmosphere for 3 days until a 100% CPE was detected in control virus wells. Serum Neutralisation Index 50 (SN₅₀) values were determined using SNT. Antibody titres were expressed as the reciprocals of the highest serum dilution resulting in the inhibition of cytopathic effects.

Results and discussion

The results of the serological investigation for the 5 viruses in the 936 cattle sera are presented in Table 1. Overall percentages of BVDV, PIV-3, BAAdV-1, BAAdV-2 and BAAdV-3 antibodies in cattle were 20.19%, 41.02%, 23.82%, 21.90% and 8.11%, respectively. Evaluation of the data revealed that 25.64% of the sampled population were seronegative against the investigated viruses and that 74.36% were seropositive for one or more them.

Results were also evaluated for single or multiple infections. Accordingly, 29.42% of the cattle were seropositive for one of the viruses, 25.42% for two viruses, 5.23% for three viruses and 0.1% for four viruses. No seropositive cattle for five viruses were detected.

The percentages of antibodies against a single virus in cattle were found 4.36%, 8.76%, 7.15%, 7.47% and 1.70% for BVDV, PIV-3 and BAAdVs-1, 2 and 3, respectively.

SN₅₀ values of BVDV, PIV-3, BAAdV-1, BAAdV-2, BAAdV-3 varied from 1 : 5 to 1 : 320, 1 : 5 to = 1 : 320, 1 : 10 to = 1 : 320, 1.10 to 1 : 20 and 1 : 10 to = 1 : 320, respectively, as detailed in table 2.

In this study, the prevalence of BVDV, PIV-3, BAAdV-1, BAAdV-2 and BAAdV-3 which cause respiratory disease in cattle was serologically analysed. General evaluation of the data revealed that 25.64% of the sampled population were seronegative against the 5 viruses and that 74.36% were seropositive for one or more of them.

Overall percentage of seropositivity against BVDV, PIV-3, BAAdV-1 BAAdV-2 and BAAdV-3 in cattle were 20.19%, 41.02%, 23.82%, 21.90% and 8.11%, respectively. The seroprevalence rates for these viruses,

Tab. 1. Distribution of seroprevalance of BVDV, PIV-3, BAAdVs 1, 2 and 3 by herd and provinces (results are showed as number of positive sera and percentage of seropositivity in parenthesis) (n = 936)

Province	Herd Code	No. of tested animals	No of seropositive animals (%)				
			BVDV	PIV-3	BAAdV-1	BAAdV-2	BAAdV-3
Samsun	A	315	69 (21.90)	96 (30.47)	92 (29.20)	119 (37.77)	32 (10.15)
	B	71	14 (19.71)	11 (15.49)	1 (0.14)	-	-
Tokat	C	107	13 (12.14)	77 (71.96)	19 (17.75)	1 (0.09)	7 (0.6)
	E	83	16 (19.27)	72 (67.28)	12 (14.45)	-	-
Sivas	F	272	44 (16.17)	81 (29.77)	26 (0.95)	-	7 (0.2)
	G	88	33 (37.50)	47 (53.40)	73 (82.95)	85 (96.59)	30 (34.09)
Total		936	189 (20.19)	384 (41.02)	223 (23.82)	205 (21.90)	76 (8.11)

Tab. 2. Antibody titer of positive sera. Antibody titers were determined with Serum Neutralisation Index (SN₅₀)

SN ₅₀ values of positive sera	Viruses (%)				
	BVDV	PIV-3	BAAdV-1	BAAdV-2	BAAdV-3
≤ 1 : 20	155 (82.01)	324 (84.37)	176 (78.9)	205 (100)	46 (60.5)
1 : 20-1 : 40	15 (7.9)	21 (5.4)	27 (12.1)	-	11 (14.4)
1 : 40-1 : 80	5 (2.6)	9 (2.3)	11 (4.9)	-	6 (7.8)
1 : 80-160	3 (1.5)	6 (1.5)	8 (3.5)	-	5 (6.5)
1 : 160-1 : 320	11 (5.82)	20 (5.2)	-	-	5 (6.5)
≥ 1 : 320	-	4 (1.04)	1 (0.4)	-	3 (3.9)
Total	189	384	223	205	76

except for BVDV, were similar to previous reports for cattle in Turkey (1, 2, 4, 5) The sampled animals had not been vaccinated against any of the mentioned viruses, indicating that the results arose from natural infections. Differences among the its BVDV data and previous studies can be explained with the investigated region and animal population. BVDV causes genital system problems and persistent infections which are economically important, as well as respiratory system infections. Therefore, the prevalence of BVDV infection in herds should be determined and strategies for its eradication should be developed. Seropositivity rates determined for PIV-3 (41.02%) can be explained by the high infectiousness of the virus in the normal environment of tested animals.

The total seropositivity rate to BAAdV-1, 2 and 3 (23.82%, 21.90% and 8.11%, respectively) varied from herd to herd. BAAdV-1 antibodies were detected in all herds. BAAdV-2 antibodies were detected in three herds while BAAdV-3 antibodies were identified in four of the herds.

Alkan et al. (2) determined 9.38% positivity against only one agent, 11.46% for two agents and 72.01% for three-eight agents in the cattle in their seroprevalance study performed on nine viruses (IBR, PI-3, BRSV, BVDV, BAAdV1, BAAdV2, BAAdV3, BEV 1 and 2). Akça et al. (1) reported 20.5% seropositivity against a single virus, 29.4% for two viruses, 27.2% for three viruses

and 9.7% for four to five viruses in their study of BAdV-1, BAdV-2, BAdV-3, BLV, BRSV and PIV-3 in buffalo. Lauchli et al. (10) studied BRSV, BAdV-1 and 4, coronavirus BVDV and PIV-3 in cattle and reported a single infection in 25% of the animals and multiple infections in 75%. In the present study, 29.48% of the animals were seropositive for only one virus, 25.42% for two viruses, 5.23% for three viruses and 0.1% for four viruses. No animals were seropositive for 5 viruses.

SN₅₀ results of the investigated sera were detected between 1 : 5 - : ≥ 320 . The highest antibody values ($\geq 1 : 160$) were calculated for BVDV, PI-3 BAdV-1 and BAdV-3. These results show evidence of recent, severe infections caused by BAdV-1, BAdV-3, BVDV and PI-3 viruses in the investigated region.

In conclusion, serological data for some viruses causing respiratory infections in a limited cattle population in 3 provinces were updated. The results indicate that it is necessary to have effective protection against BVDV and PIV-3 infections. It is recommended that cattle in rural areas be vaccinated.

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